

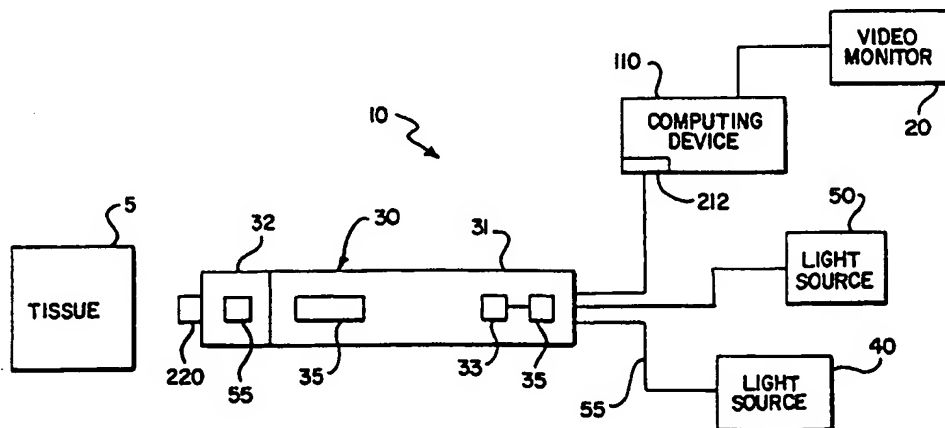
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(54) Title: IMAGING DISEASED TISSUE USING AUTOFLUORESCENCE



## (57) Abstract

An apparatus for imaging body tissue utilizing autofluorescence includes an endoscope (30) having a distal end (32), proximal end (31) and a light processing unit (33) located at the proximal end (31). The light processing unit accepts light in a spectral region from a light source (40) and directs the light through the endoscope to the tissue (5) located at the distal end (32). The light excites autofluorescence in the tissue. Optics (35) located in the distal end (32) of the endoscope collect the excited autofluorescence in a spatially resolved manner and routes it to the light processing unit (33) which separates the emitted light from any reflected excitation light. A plurality of imaging detectors (45) receive the emitted light. Each imaging detector is responsive to light in a different spectral band. An apparatus for sampling tissue includes an endoscopic device (30) comprising a sampling device (220) capable of taking a plurality of tissue samples, deposited in separate captive units (226).

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## IMAGING DISEASED TISSUE USING AUTOFLUORESCENCE

**BACKGROUND OF THE INVENTION**5    **A.     Field of the Invention**

The present invention relates to an apparatus and method for imaging bodily tissue by using autofluorescence. More particularly, the present invention relates to an endoscopic apparatus and method for imaging and sampling bodily tissue using autofluorescence techniques.

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**B.     Description of Related Art**

Cervical cancer often begins as a precancerous lesion on the cervix (i.e., the outer end of the uterus) and is called cervical intraepithelial neoplasia (CIN). The lesion can deepen over a period of years and if left untreated can become an invasive cancer. A Pap  
15    smear test is currently a common method of providing a type of screening for cervical cancer. The test involves taking a sample of cells from the cervix, and sending the sample to a laboratory to be analyzed. Test results usually take two or three weeks to complete.

If the laboratory analysis determines that abnormal cells are detected from a first Pap test, a follow up test is typically performed. A second abnormal Pap smear will often  
20    prompt a colposcopic examination wherein the cervix is examined usually with a low-power stereo microscope. During colposcopy, suspect abnormal tissue is often biopsied and again sent to a laboratory for analysis. Because patients must often wait another two to three weeks for these results, a heightened period of anxiety and fear for the women and their families is created. Often, the first and second abnormal Pap smear result from false  
25    positive test results. Therefore, oftentimes, when a tissue sample has been biopsied, the sampled tissue was incorrectly determined to be cancerous and did not need to be removed.

Spectroscopic autofluorescence, a minimally invasive procedure for analysis of cervical cytological, has been used to decrease a number of the problems normally associated with Pap smear tests.

Spectroscopic methods for differentiating cervical neoplasia from normal cervical tissue *in vivo* can be used to detect abnormal cells on the outside of the cervix. Typically, a fluorescence spectroscopy has optical fibers at the end of a small probe which illuminate areas of the cervix. Suspect tissue is exposed to ultraviolet and visible laser or lamp light, causing substances naturally present in the tissue to fluoresce. The specific wavelength or signature of the light absorbed and emitted by cervical tissue is analyzed. The fluorescence spectra is then measured and compared at different intensities and wavelengths since abnormal or cancerous tissues consistently display different results from normal or non-cancerous tissue. Typically, a computer algorithm analyzes the fluorescence spectrum and assesses the degree of cell abnormality.

Generally, there are two types of fluorescence measurement techniques: the first being emission spectroscopy and the second being excitation spectroscopy. In emission spectroscopy, the exciting light is kept at a fixed wavelength and the emitted fluorescent intensity is measured as a function of the emitted wavelength. In excitation spectroscopy, the emission wavelength is kept fixed and the fluorescence intensity is measured as a function of the excitation wavelength.

Both emission and excitation spectra measurements have limitations. For example, both types of spectra measurements analyze only a single parameter to determine cell abnormality. The nature of the human tissue, however, is such that the application of any one single method produces a large amount of data, most of which is extraneous to the intended measurement. A primary reason for this situation is that tissues contain an extensive and diverse assortment of fluorescent species. Many of the species are present in

high concentrations and have excitation bands distributed throughout the ultraviolet and the visible spectra regions.

Another limitation is that the emission band of one fluorophore may overlap the excitation band of another fluorophore, consequently leading to energy transfer between the emission and excitation bands. Consequently, emissions from one fluorophore could possibly excite another fluorophore. The net effect is that optically exciting a tissue sample at almost any wavelength in the ultraviolet or visible wavelength regions causes tissue autofluorescence over a broad spectral range. As these emissions are typically composed of contributions from multiple fluorophores, utilizing a single analytical parameter makes the autofluorescence spectrum complex and problematic to solve. Consequently, a robust discrimination between tissue states is often difficult to obtain.

Another limitation of typical fluorescence measurement techniques is that they cannot be readily combined with an apparatus or method for taking a biopsy. In other words, once an abnormal tissue area is detected, samples from this particular suspect area cannot be simply, quickly and accurately taken. With current devices utilizing fluorescent measurement techniques, after locating the abnormal area, the endoscope must be withdrawn from the patient. Once the endoscope is withdrawn, the Pap smear specimen can then be taken as a blind sample. Typically, there is no correlation between where on the cervix the sample is taken and where the suspect tissue was identified. Taking a blind sample, therefore, often results in samples being taken from normal areas or perhaps even areas which have not been previously investigated. Usually, the sample is also taken by relatively imprecise sampling devices such as brushes, scraper devices or the like.

Once the sample has been taken and withdrawn from the body, the specimen is typically smeared onto a microscope slide. This is often done by the physician performing the test. The slide is then submitted to a remote laboratory for cytopathological microscopic

examination. Pertinent patient data must be sent along with the slide including the medical history, day in menstrual cycle, family history and other known risk factors. Gathering and collating these patient data, which are critical to the proper evaluation of a specimen, is a time-consuming, expensive, inefficient and labor-intensive process. The laboratory administrative personnel who gather such data are also responsible for manually recording the results of the Pap smear tests and ensuring that both the slide and paperwork provided to the cytotechnologist relate to the same patient. As the complexity of testing, analyzing, handling and transporting the Pap smear samples increases, the probability for a false positive, a false negative or sample contamination increases.

10       The typical Pap smear test has a number of other disadvantages. For example, in almost every instance where a slide specimen is produced, the slide is forwarded to a laboratory. No preliminary analysis to eliminate possible unnecessary laboratory testing is conducted. This increases the cost of performing a Pap smear since there is no preliminary determination as to the possibility of normality or absence of abnormality. Moreover, 15 because the sampling is taken "blind", there is typically no assurance as to whether the suspect abnormal cells have in fact been sampled. Oftentimes, only after having forwarded the sample to the testing facility and waiting two to three weeks is it eventually determined that another sample must be taken. Incidents of poor sample or slide preparation are also common because of the large amount of human interface with each specimen slide. 20 Moreover, because slides are often sent to a location remote, there is an increased risk that the sample may become lost, broken or contaminated. The complexity of maintaining a secure and sterile transporting medium further increases the cost of sample transport. In addition, there is a psychological disadvantage in having to wait up to two weeks or longer for the test results to either confirm or rebut a primary abnormal reading.

### SUMMARY OF THE INVENTION

The present invention relates to an apparatus and method for imaging body tissue utilizing autofluorescence. The apparatus includes an endoscope having a distal and proximal end and a light processing unit located at the proximal end of the endoscope. The light processing unit accepts light in a spectral region from a light source and directs the light through the endoscope to the tissue located adjacent the distal end of the endoscope. The light excites autofluorescence in the tissue. Optics located in the distal end of the endoscope collect the excited autofluorescence in a spatially resolved manner and routes it to the light processing unit which processes the emitted light from any reflected excitation light. Imaging detectors receive the emitted light. Each imaging detector is responsive to light in a different spectral band and each generates a separate image. An image is generated according to a plurality of independent measurements.

In another aspect of the invention, an apparatus for sampling tissue includes an endoscope having a sampling device. The sampling device takes a plurality of tissue samples. Captive units are in communication with the sampling device such that each tissue sample is deposited in a separate captive unit.

These and many other features and advantages of the invention will become more apparent from the following detailed description of the preferred embodiments of the invention.

**BRIEF DESCRIPTION OF THE DRAWINGS**

FIG. 1 is a schematic view of an imaging video endoscopic system incorporating a preferred embodiment of the present invention.

FIG. 2 is a schematic view of the endoscope shown in FIG. 1.

5        FIG. 3 is a data input block flowchart for generating a preprocessed image.

FIG. 4 illustrates a ratio block flowchart for generating a composite ratio image from the data input blocks generated by the flowchart shown in FIG. 3.

FIG. 5 is a schematic view of an alternative embodiment of the endoscope shown in FIG. 1.

10        FIG. 6 is a schematic view of an imaging video endoscope system incorporating another preferred embodiment of the present invention.



### DETAILED DESCRIPTION OF THE PREFERRED EMBODIMENTS

FIG. 1 illustrates a video endoscopic system 10 incorporating a preferred embodiment of the present invention. The system 10 is utilized as a diagnostic tool for imaging tissue 5. Preferably, the tissue 5 being investigated by the endoscopic system 10 is the tissue of a cervix. Alternatively, the endoscopic system 10 is a diagnostic tool used to generate an image of any *in vivo* tissue where use of minimally invasive procedures are advantageous. The endoscopic system 10 utilizes the autofluorescence and reflectance properties of tissues to discriminate between normal and abnormal tissues.

The system 10 includes an endoscope 30, a video monitor 20, a data collection device or computing device 110 and a light source 40. Alternatively, a second light source 50 is provided and is preferably a white light source. The imaging endoscopic system 10 is suitably sized and shaped for cervical inspection. The endoscope 30, having a distal end 32 and proximal end 31, includes a light processing unit 33, distal optics 35, and imaging detectors 45. Preferably, the light processing unit 33 is either a wavelength division multiplexer or a beam splitter. As will be further discussed with reference to FIG. 2, the endoscope 30 also has a sampling device 220.

The endoscope 30 functions as an imaging reflectance fluorometer or as a reflectance spectrometer depending upon whether the light processing unit 33 is a wavelength division multiplexer or a light processing unit beamsplitter, respectively. The light processing unit 33 accepts light in a spectral region which is generated by the light source 40. The light generated by the light source 40 is communicated to the light processing unit by way of fiber optic cable 55. Alternatively, the endoscope includes both a wavelength division multiplexer and a beam splitter may be concatenated to provide both functional modes simultaneously.

The light generated by the light source 40 or 50 is communicated to the light processing unit 33 by way of fiber optic cable 55. The light emitted by light source 40, 50 may encompass a continuum of wavelengths within the ultraviolet, visible and near infrared spectral regions or one or more wavelengths or groups of wavelengths within one or more  
5 of these regions. The light processing unit 33, which is used for fluorescence or reflectance measurements, respectively, accepts light of the appropriate spectral distribution from either the first light source 40 or the second light source 50 by way of the fiber optic cable 55. The light is directed from the light processing unit 33 through the endoscope 30 to the distal end 32 where the light illuminates the tissue 5. The endoscope 30 includes optical fibers  
10 which can transmit light in both directions between the distal end 32 and the proximal end 31.

The light emerging from the distal end 32 of the endoscope 30 is reflected by and excites autofluorescence in the tissue 5. The light reflected by and the autofluorescent emissions from the tissue 5 is collected by the distal optics 35 contained within the distal  
15 end 32 of the endoscope 30. Preferably, this light is collected by the distal optics 35 and transmitted to the proximal end 31 of the scope in a spatially resolved manner where it is routed to the light processing unit 33. The light processing unit 33 separates the light reflected from or emitted by the tissue 5 from the light 41 being directed from either light source 40 or 50 to the proximal end 31 of the scope. The light processing unit further  
20 separates light emitted from the tissue 5 from light reflected by the tissue 5. The light emerging from the light processing unit 33 is routed to one or more imaging detectors 45. Each of the imaging detectors 45 or fluorescence detectors 45 is responsive to light in a different spectral band. Each detector 45 generates a separate image representative of the specific spectral band associated with that detector 45.

Each generated image is communicated to a computing device 110 for manipulating and storing the images. Preferably, the computing device 110 is in communication with a video monitor 20 wherein the images, either individually or as a composite of a plurality of images can, after the appropriate manipulations, be examined for possible tissue abnormalities.

In a combined image, due to the difference in the wavelengths of autofluorescence between normal and abnormal tissues, various regions of the tissue 5 having abnormalities are visible. In the preferred embodiment where a cervix is being examined, the composite image facilitates localization of any abnormalities on the surface of the cervix. The computing device 110 can alternatively perform image processing and image analysis techniques such as edge enhancement and segmentation which can be applied to these images either singly or in combination thereby facilitating abnormality detection and interpretation. Feature recognition permits electronically establishing visual reference points on the examined tissue and relates to the location of features of interest to these reference points. Furthermore, this capability allows joining multiple contiguous fields of view to produce a panoramic display. Once an area of abnormality in the tissue 5 is detected, the scope 30 may then take a sample of this area.

Information relating to the state of the tissue 5 can be obtained through various methods. For example, in one method of differentiating between normal and abnormal tissues, the intensities of flavinoid autofluorescence at several selected wavelengths is measured. The ratios between these emission intensities for normal and abnormal tissues vary in a characteristic manner. By varying the light transmitted by the light processing device 33 and therefore the excitation wavelengths, various other cellular constituents such as porphyrins can be made to autofluorescence in a diagnostically useful manner, and similarly characteristic ratios can be computed. Computing such a ratio on a pixel by pixel

basis from a suitably selected pair of images can produce a derived ratiometric image in which the differences between normal and abnormal tissue are accentuated. These derived images may be further processed by methods such as edge enhancement and segmentation to further accentuate any differentiation.

5        It is generally known that one can differentiate between normal and abnormal tissues by exciting autofluorescence by illuminating the tissue with light in one wavelength band, measuring the intensity of the light emitted in one or more wavelength bands, computing ratios between those emitted intensities, and discriminating between normality and abnormality on the basis of these ratios. However, since tissue autofluorescence is  
10 comprised of emissive contributions from a multiplicity of fluorophores and, even under ideal conditions, the emissions from a single fluorophore tends to be spectrally broad, the autofluorescence spectrum of tissue tends to be relatively undifferentiated with few pronounced features. Furthermore, the fluorescent emission intensities at multiple wavelengths under a single excitation condition are highly correlated. Therefore, the  
15 information gained by computing intensity ratios between multiple pairs of emission wavelengths represents only an incremental improvement over that obtained from computing the ratio between a single pair of emission wavelengths.

In a preferred embodiment, the system 10 utilizes information obtained at a multiplicity of emission wavelengths generated at a multiplicity of excitation wavelengths,  
20 each wavelength combination selected to, in and of itself, to maximize discriminatory power between normal and abnormal tissues. Therefore, instead of relying upon a single measurement to obtain the desired differentiation, system 10 utilizes multiple independent measurements that are combined to obtain substantially improved discrimination. Furthermore, as described below, system 10 allows the results obtained through the use of  
25 multiple independent measurement techniques to be combined to further improve the

robustness of the discrimination between normal and abnormal tissues. The use of statistically based classification functions and multi-dimensional pattern matching techniques to effect this merging and interpretation on the multiplicity of independent data sets furthers this goal.

5        Preferably, additional information is derived and interpreted by acquiring the autofluorescence signals in a time resolved manner. The relaxation times and fluorescent lifetimes of different fluorophores, which are determined from time resolved measurements, differ substantially between fluorophores. The various relaxation times can therefore provide an indication of a fluorophore's identity. These parameters are frequently influenced  
10 by the environment surrounding the fluorophore in ways that, in turn, reflect the normality or abnormality of the surrounding tissue.

In another preferred embodiment, reflectance spectrometry provides yet another means of probing tissue status as changes in the tissue status are often evidenced by the changes in colored constituents of the tissue. Although normally practiced in the visual  
15 spectral region, reflectance spectrometry can be extended into the near infra red as well as ultraviolet. Reflectance spectrometry is extended to a depth where the incident light penetrates the tissue sufficient enough such that additional tissue information can be obtained. This information could include such characteristics as the concentrations of certain metabolites and the degree of blood oxygenation. Raman scattering could also be  
20 used because of increased incident light penetration and the fact that there is a relative scarcity of fluorophores having excitation bands in the near infra red.

The previously discussed methods can be used individually as a means of detecting, and in some cases interpreting differences in tissue status. Unfortunately, the nature of human tissue is such that the application of any one of these methods produces an  
25 overabundance of data, most of which is extraneous to the intended measurement. The

primary contributor to this situation is that tissues contain an extensive and diverse assortment of fluorescent species. Many of the fluorescent species are present in high concentrations and have excitation bands distributed throughout the ultraviolet and most of the visible spectral regions.

5       As previously discussed, the emission band of one fluorophore may overlap the excitation band of another fluorophore thereby leading to energy transfer between the two. Consequently, there is a distinct possibility that emissions from one fluorophore may excite another. The net effect is that optically exciting a tissue sample having a wavelength in the ultraviolet or visible wavelength region will cause the tissue to autofluorescence over a  
10   broad spectral range. As these emissions typically are composed of contributions from multiple fluorophores, the autofluorescence spectrum is complex and difficult to resolve. This, in turn, makes it difficult to obtain a robust discrimination between tissue states through the use of a single method.

A preferred embodiment of the present invention resolves these problems by  
15   applying methods that have been developed for applications such as the mapping of natural resources and military reconnaissance. These "multispectral" methods in effect "fuse" or combine the outputs of multiple sensing modalities to obtain a result that is considerably more robust than the results obtained using any one modality independently.

For example, the classical approach to determining tissue autofluorescence is to treat  
20   the tissue as a single homogenous entity, excite it at one wavelength and measure the emissions at another wavelength. This sort of measurement is critically dependent upon using a stable, well-calibrated instrument and upon having negligible, or at least relatively low extraneous background fluorescence at the emitted wavelength. In contrast to the classical approach, robust determinations are preferably made by measuring the  
25   fluorescence at two or more emission wavelengths. The use of multiple emission

wavelengths performs internal consistency checks. Calculating ratios between the emission intensities provides another means for discriminating between changes of interest from background noise.

Because the classical approach treats the tissue 5 as a single homogeneous entity, the classical approach does not provide the spatial resolution needed to determine whether an abnormality is localized or whether an abnormality is widely distributed. Generating the fluorescence measurements described above at multiple discrete points on an image provides the spatial information needed to determine the location of any abnormalities.

In an alternative embodiment, multiple excitation wavelengths are used and other techniques are applied such as time resolved spectrometry. Each additional parameter applied supplies unique information that can be used to discriminate between tissue states. However, the multiple parameters provide redundant or extraneous information. Adding derived parameters such as intensity ratios to the data set can define or reflect significant tissue characteristics that can, in turn, facilitate interpretation.

The preferred approach to interpreting this voluminous mass of data is "preponderance of evidence" wherein each data set is interpreted by the computing device independently of any other data set. A majority vote method is applied to the collection of conclusions derived from the data sets. An alternative, more sophisticated approach applies a multivariate classifier or discriminate function that combines information from the various data sets in a prescribed, and statistical manner. A single composite result is then obtained. Such methods are usually applied at each point or pixel of an image in a spatially resolved image. Incorporating multiple lines of evidence into a single determination works to filter out extraneous and redundant information while improving the robustness of the final determination.

In another preferred embodiment, more advanced techniques such as contour following are used. For example, if the data sets are viewed as a stack of image planes each of which collected under different defined conditions, a contour through the stack describes the changes or evolution of the signal level at each pixel as a function of the measurement  
5 parameters. The shape or shapes of the contours provide additional discrimination between different possible interpretations.

The endoscopic system 10 of FIG. 1 acquires data under a multiplicity of conditions as previously described. The computing device 110 evaluates the image data acquired under these conditions. The generated data is interpreted, not in isolation of each individual  
10 generated parameter, but in its entirety as a composite whole. This preferred composite analysis improves the accuracy and robustness of the final determination as to the normality or abnormality of the tissue 5. Due to the large amounts of data involved in this preferred composite type of approach, it may be desirable to apply fuzzy logic. Alternatively, a neural network or other "self-teaching" method can be used for the interpretation of either  
15 the entire data set or to subsets thereof.

In addition to the individual images taken under multiple conditions, the second light source 50 is used to generate a "white light" image by directly using broad band illumination. Alternatively, the white light image is synthesized from multiple narrow band images.

20 In addition to its potential relating to its diagnostic utility, the white light image allows identifying visual positional reference features within the field of view of the endoscope. In a preferred embodiment, the white light image is overlaid with markers. The markers designate certain reference features, locations or regions of the tissue that analytical methods such as those described above have identified as being abnormal or suspect. Such  
25 reference features facilitate the ability of an examining physician to return at a later time to



a previously investigated location of the cervix. For example, if the physician performs a preliminary test to evaluate a sample recently taken and subsequently determines that, for one reason or another, a follow-up sample is required, the location from where the initial sample was taken can be quickly and accurately identified.

5        Preferably, two types of interpreting schemes are used to interpret the multiplicity of data sets generated by the system 10. Before these interpreting schemes are described, however, the processing of the generated data sets will be discussed.

FIG. 3 illustrates a data input block flow chart 300 which shows how a preferred embodiment of the present invention generates a preprocessed image 310. First, image data  
10    305 is acquired at the image acquiring step 305. The image data relates to a particular excitation wavelength N and a particular emission wavelength M which together define an initial image NM. Once the image data and therefore the initial image NM is acquired, shading correction is applied during a shading correction step 307. After the image has been corrected for shading, curvature correction is applied during a curvature correction  
15    step 309. The preprocessed image NM is then defined as an image block NM. Various image blocks can be similarly generated for each excitation wavelength N and each emission wavelength M pair. Preferably, a plurality of image blocks NM are generated and configured as data input blocks, which in turn are used to generate a composite image. A resulting preprocessed image NM 310 is then used generate a plurality of ratio blocks as  
20    shown in FIG. 4.

FIG. 4 illustrates a ratio block flow chart 400 for generating a composite ratio image from the data input blocks 320 generated in FIG. 3. First, the ratio block flowchart 400 generates a plurality of intensity ratios 410 during the intensity ratio step. An intensity ratio  
415 is computed for varying data input blocks 320. For example, a first intensity ratio Ratio  
25    NM12 415 is computed for the data input blocks NM1 and NM2. A second intensity ratio

Ratio NM23 417 is computed for the data input blocks NM2 and NM3. This process is repeated for each different emission wavelength M. Preferably, an intensity ratio is computed for each pixel of the resulting image. The generation of intensity ratios are repeated for each pair of excitation wavelengths N and emission wavelengths of interest M.

5 Preferably, at least two emission wavelengths M for each excitation wavelength are generated. The resulting matrix of generated intensity ratios is then used to generate a ratio image which is then interpreted to analyze the state of the tissue.

In a preferred embodiment, a first approach to interpreting the ratio image applies a first and a second threshold value to each pixel in the ratio image. The ratio image is then

10 segmented, categorized or defined into various regions. Preferably, the pixels of the ratio image are segmented into either normal, suspect or abnormal regions. Preferably, the first and the second threshold values are empirically derived based upon the statistical distribution of ratio values in a large number of reference images. For example, an upper (Tu) and a lower (Tb) threshold are defined and are used for image interpretation.

15 For example, if a ratio value at a given pixel position is defined as R, then three scenarios are possible. First, if the ratio value R is greater than the upper threshold Tu, then the pixel is classified as being abnormal. Second, if the ratio value R is greater than the lower threshold Tb but less than the upper threshold Tu, then the pixel is classified as being suspect. Third, if ratio value R is less than the lower threshold Tb, then the pixel is

20 classified as being normal. A resulting image can then be generated and analyzed according to pixel classification.

In another preferred embodiment, a segmentation algorithm is applied to interpret the ratio image. First, a search is made of the entire ratio image for local minima. Alternatively, a search is made of the entire ratio image for local maxima. In the case where

25 a search for local minima is made, at each local minimum found, a temporary threshold

value is defined and equated to the ratio value at the minimum plus one. The ratio values at all neighboring pixel locations are then examined. If the value at the neighboring pixel being examined is less than or equal to the temporary threshold value, the neighboring pixel is tagged with the identifier for the minimum with which it is now associated. If the pixel  
5 has been previously tagged, the designation is not changed. This cycle is repeated until all pixels making up the ratio image are "tagged." All pixels having the same tag value are considered to belong to the same region, whether that region is defined as normal, abnormal or suspect.

Preferably, a conventional matched filter is applied to detect particular features or  
10 feature shapes within the initial preprocessed image or the ratio image or alternatively in the threshold renditions of these images. Filtering, usually in combination with dilation and erosion, smoothes the boundaries between regions and removes noise from the resulting images.

The preprocessed images generated as described in FIG. 3 or the ratio image  
15 generated as described with reference to FIG. 4 can also be interpreted using a linear or statistical classifier. Preferably, the classifiers are of the form: Figure of Merit =  $F(I_1, I_2, I_3, \dots, I_n)$  where  $I$  represents pixel values in the preprocessed image or the ratio image. Preferably, the function "F" is determined by the multivariate statistical analysis of a large population of reference images. Standard statistical tests for significance determine which  
20 images or pixels are used in interpreting measurements made on any particular type of tissue image. The multivariate statistical analysis effectively determines how much weight should be given to each of the remaining parameters.

FIG. 2 illustrates the details of the endoscope 30 shown in FIG. 1. The endoscope  
30 may be rigid or flexible. Endoscope 30 includes a sheathing member 205, a distal end or  
25 endoscope tip 32, and a sampling device 220 located at the distal end or tip 32. The

sampling device 220 is used to extract samples from the tissue 5 under investigation. Preferably, the sampling device 220 is manipulated by a physician while performing autofluorescence imaging as previously discussed. The sampling device 220 is also steerable. By steerable, it is meant that the sampling device 220 is rotatable, pivotable and retractable within the endoscope sheathing 205 in such a manner that the sheathing 205 does not need to be manipulated or controlled.

Preferably, the sampling device 220 is configured as a brush 225 having a plurality of sampling members 226. To take a sample of tissue 5, the sampling device 220 is manipulated and steered to a location adjacent the suspect tissue 5. The brush 225 rotates preferably in a counter-clockwise direction. The sampling device 220 or more preferably the brush 225 extends towards the tissue 5 such that the sampling members 226 come into contact with the tissue 5. The rotating sampling members 225 securely remove an outer cell layer of the examined tissue 5.

Alternatively, the sampling device 220 includes a sampling ribbon, an unwind reel and a rewind reel. Before any samples have been taken, the entire sampling ribbon resides on the unwind reel. As samples are taken, a predetermined length of the sampling ribbon is unwound from the unwind reel onto a rewind reel. During sampling, a portion of the ribbon comes into contact with the tissue 5 thereby securing a tissue sample. As subsequent samples are taken, the rewind reel takes up the sampling ribbon segment containing the sampled tissue. Another sample can then be taken. Once the entire sampling ribbon is completely transferred from the unwind to the rewind reel, the sampled tissue stored on the ribbon of the rewind reel can be taken out of the endoscope 30 and tested. The sampling ribbon can then be tested at the physician's facility and then sent to a laboratory for further testing.

During the imaging procedure previously described with respect to FIG. 1, the sampling device 220 remains in a retracted state. In this retracted state, the sampling device 220 remains inside the sheathing 205 of the endoscope 200 and decreases any interference the sampling device 220 may create during imaging. Once the physician operating the scope 5 30 determines that a sample of the investigated tissue 5 should be taken, the sampling device 220 is extended beyond the distal end of the scope 30, towards the tissue 5. After a sample is taken, the sampling device 220 can then be retracted back within the distal end 206 of the endoscope 200. Further imaging or sampling can then take place.

Preferably, the sampling device 220 removes a plurality of tissue samples from the 10 tissue 5 in a sequential order. This enables the operator of the device to collect a set of samples from either the same suspect location or alternatively from a variety of different areas. The ability to take a plurality of tissue samples during one minimally invasive procedure results in a number of benefits. For example, where a plurality of samples are taken from the same or different location, specimens will generally have an enhanced 15 probability of containing abnormal cells. Moreover, having a collection of samples taken from an abnormal tissue area increases the probability that the samples contain a portion of the suspect tissue which initially gave rise to the determination that a sample should be collected. Having a set of samples that are not taken blindly also enables the physician to revisit and perform further investigation of various, previously investigated suspect areas.

20 Preferably, once the sampling device 220 extracts a sample, the sample is drawn into a captive unit 260. Preferably, the captive unit 260 is a sterile container such as a sleeve, capsule, or like device. More preferably, the endoscope 30 contains a plurality of captive units 260 such that each time a tissue sample is taken, the sample is placed in its own, separate sterile captive device 260. The captive device 260 can be provided with an 25 identification means such as a label, print-out, log or other similar type of identifier. The

captive device 260 is preferably detachable from the sampling device 220 and therefore detachable from the endoscope 30. Therefore, the captive units 260 can be used as storage or shipping containers. The storage container simplifies the transporting, marking and identifying various aspects of the sample.

- 5            Preferably, the captive units 260 are disposable. After final testing of a sample, a captive unit 260 originally containing the sample can therefore be disposed of. Alternatively, the captive unit 260 is reusable such that it can be repeatedly sterilized and reused.

            The extracted samples contained within the captive units 260 can be initially  
10    examined at the physician's location and then subsequently sent to a laboratory for further testing. Preferably, the physician performs an initial test on the sample. The initial test can be used to determine whether the samples are indeed samples taken from the investigated suspect tissue. The initial test also enables the physician to make a relatively quick determination as to whether any additional samples of the patient are required. Local  
15    testing also enables the physician to determine relatively quickly whether the tissue is actually abnormal. This is an important consideration since it has been documented that over ninety percent of Pap smear tests sent to labs for testing result in a negative result. Consequently, by providing the physician with a preliminary screening test for determining whether the sample contains abnormalities provides a number of advantages.

- 20            For example, the patient is not required to go through an additional two or three weeks of anxiety waiting for the results of the test. The samples removed by way of the previously discussed method can also be examined by any standard method. The initial test is performed by the physician by taking a portion of the sample tissue and smearing it on a slide. This slide can then be analyzed at the same location where the sample was taken. If  
25    this preliminary test or screen performed by the physician results in an abnormal reading,

the entire captive unit can then be transported to a laboratory where the standard analysis can be conducted.

By being able to take a plurality of samples, a "map" of cervix sampling locations is generated. By mapping previously sampled locations and by using the previously discussed  
5 visual reference points, these same sampling locations can be revisited during subsequent follow-up examinations. In addition, by taking a plurality of tissue samples and thereby enhancing the probability of detecting and collecting abnormal cells, the proposed system also provides a cost effective and efficient means for follow up testing based on an abnormal Pap smear.

10 One preferred means of performing an immediate examination of a sample is to subject the sample to fluorimetric measurements similar to those described above. In this preferred method, an aliquot of the sample taken by the sampling device may be tested in its current state (i.e., "as is"). Alternatively, the sample is suspended within a captive unit in an appropriate fluid medium. In this alternative embodiment, the sampling device places each  
15 sample into a captive unit. The unit holds both the extracted sample and a fluid medium. This method eliminates a number of the problematic areas normally associated with the preparation, transportation, and testing of Pap test samples. Moreover, this method reduces the number of samples that are sent out for costly analysis to an off-site investigation. In addition, because the number of Pap smear false positive test results will be decreased,  
20 anxiety of women waiting for follow up test results will be decreased.

As the conditions surrounding the proposed methods of Pap test re-testing are better defined and controlled than those used for the *in-situ* identification of sampling regions, a positive second test provides a high probability of confirming that the sampled tissue was abnormal. This information can then be used as a means of verifying that the intended

sample was indeed taken and could be used to determine which samples should be subjected to a more vigorous analysis.

Return to FIG. 1, the present system 10 may utilize both intrinsic and extrinsic testing. Therefore, although it is not essential for using the system 10, one alternative  
5 embodiment of the present invention utilizes photodynamic therapy. During such extrinsic testing, a photodynamic agent or photodynamic drug is applied to tissue to be examined. The drug will then generally be incorporated into atypical or cancerous cells.

In some circumstances, the system 10 utilizes a drug to label atypical or cancerous cells. However, the system 10 may also use other suitable probes that assist in the  
10 identification of atypical or cancerous cells. Applicants note, however, that the use of probes may often provide advantages.

With photodynamic therapy ("PDT"), a PDT drug is a probe that can label abnormal tissue in the examined area. This labeling occurs when the photodynamic drug is metabolically incorporated into an atypical or cancerous cell in a substantially higher (or  
15 simply different) concentration than typical or non-cancerous cells.

Tests that rely upon the metabolic incorporation of a drug into a cell provides a number of advantages over tissue analysis with autofluorescence alone. For example, by metabolically incorporating a drug into a cell or tissue region, atypical cells or atypical tissue regions can be made to fluoresce. Induced fluorescence, caused by excitation with an  
20 illumination or light source, results in increased sensitivity and increased specificity, compared with visual examination of the tissue.

The photodynamic therapy drug or agent ("agent") may be administered in a number of different methods. Typically, the agent will require to be applied a predetermined time period before the subject area can be investigated. For instance, often after application of an  
25 agent, the agent may take 60 minutes or longer to be metabolically incorporated into *in vivo*



tissue. This somewhat lengthy incorporation time period may result, for example, in a patient having to wait an hour or more before an *in-vivo* examination may commence.

To reduce or eliminate the need for such waiting period while in the physician's office (e.g., an Obstetrician/Gynecologist's practice), the agent may be administered before the patient arrives for the examination. For instance, the drug may be self-administered by the patient. Self-administration can occur by way of a number of different methods. For example, a tampon or cervical sponge may be used. Prior to the date and time of the examination, the patient may be sent a tampon or cervical sponge containing the agent. The patient can then insert the tampon or sponge into her gynecological tract a predetermined period of time prior to an examination. Alternatively, where high agent concentrations may be necessary, the tending physician may require or suggest that the agent be applied by the physician or other experienced administrator prior to the examination.

Accordingly, the agent may be applied a sufficient amount of time before cervical examination. In this way, the agent gradually reaches an adequate level of metabolic incorporation. Once an adequate metabolic level is reached, the agent can thereby provide sufficient sensitivity for discrimination between normal and abnormal tissue.

Such a procedure for applying the testing agent may also reduce the amount of time that a woman must wait in the examination room or waiting room. Additionally, this application method has psychological advantages. For example, the woman may apply the agent in the privacy and comforts of her own home. Furthermore, the concentration levels of the agent, as well as the tissue application sites of the agent, can be monitored and maintained for safety and testing efficiency.

Preferably, once the agent has been applied and has had a sufficient amount of time to induce fluorescence, which time is generally correlated to the degree of tissue atypicality, the fluorescing areas may be examined by introducing an endoscope into the

gynecological tract. The endoscope 30 may be either flexible or rigid. In either case, the endoscope allows a physician or other operator to (1) image the tissue, (2) induce florescence, (3) provide image data to an analytical instrument, including a computer system, and (4) collect site-specific tissue samples.

5       The computer system can include the computing device 110 and video monitor 20 as shown in the diagnostic tool illustrated in FIG. 1. Alternatively, the computer system can comprise the system shown in FIG. 6. The computer system 204 of FIG. 6 includes an endoscope 214, an optical fiber cable 224, a ratio unit 234, amplifiers 234, and computer unit 244. The ratio unit 234 performs the wavelength analysis as previously described. The  
10       computer unit 244 includes a monitor 245. The endoscope 214 shown in FIG. 6 may be the endoscope 30 shown in FIG. 1 or in FIG. 5.

Photodynamic therapy ("PDT") offers a number of advantages over other methods. Drugs may be more sensitive and more specific to atypical cells or cancerous cells. Thus, for example, the use of PDT drugs may result in the earlier detection of lesions in the tissue.  
15       This results from an increase in sensitivity to atypical cells. Earlier detection and consequently an earlier treatment of cancerous tissue may be possible.

In addition, by increasing the sensitivity to atypical cells, and if the test results in a high degree of specificity, unnecessary Pap tests may be avoided. Therefore, the costs and associated inconveniences of Pap testing may also be avoided in the clearly negative case.

20       Use of PDT agents also provides psychological advantages to patients undergoing Pap smear testing. For example, a Pap test result typically takes two weeks to process at a remote, clinical diagnostic cytopathology laboratory. By avoiding the need for a Pap test, the patient is not burdened, for example, with the unpleasant prospect of waiting two weeks after an initial tissue investigation to learn whether or not she has been diagnosed as having  
25       cancer. Many patients may prefer not having to wait for weeks in order to learn the results

of their Pap tests. Another advantage of PDT agents is that the PDT drugs are generally not accumulated by the human body since the drugs are generally either metabolically decomposed or are excreted by way of the natural human body functions..

As will be explained, the system 10 may obtain greater sensitivity and specificity by capturing either (substantially) point or regional measurements of examined tissue. The investigating physician may therefore obtain precise data on atypical or cancerous cells and the examined tissue's location.

Furthermore, the system 10 or 204 utilizes a single wavelength of light to monitor florescence, searching for a high (or low) reflection of a particular wavelength. Such reflection is correlated to atypical or cancerous tissue. Alternatively, the system analyzes two or more wavelengths of reflected light to determine a ratio of the different wavelength reflections. The ratio unit 234 shown in system 204 may perform such function. By analyzing two or more wavelengths, the present invention determines whether a particular tissue is atypical or cancerous.

In the exemplary embodiment shown in FIG. 6, the computing device 244 receives the florescence signals representing reflected light from within the gynecological tract. The computer computes the ratio of two different wavelengths flouresced by the illuminated tissue under investigation by the endoscope 214. The ratio is computed to determine whether a particular area of tissue under consideration in the gynecological tract is atypical or cancerous. In one preferred embodiment, the computer computes and analyzes the ratio of two different wavelengths where the compared wavelengths are (a) 400 to 500 micrometers and (b) 300 to 400 micrometers. Such an analysis compares the level of (a) Flavinoids to (b) Collagen in the examined tissue.

In an alternative embodiment of the present invention, the system performs a more complex analysis of the reflections of a plurality of wavelengths. For example, a function

weighted by the strength of reflection of one or more wavelengths, together with one or more different ratios of the levels reflections of different wavelengths, might be used to obtain a result with an even higher correlation to tissue atypicality in the gynecological tract.

5       The present invention may be used with a variety of photodynamic drugs. One such drug is Levulan. This photodynamic drug is distributed under the trademark, Levulan (5-aminolevulinic acid), and is manufactured by Dusa Pharmaceuticals, Inc., ("Dusa") of Toronto, Ontario. The drug is currently being examined for use in detecting for bladder cancer.

10       Dusa has announced that its has filed an Investigational New Drug application with the U.S. Food and Drug Administration for beginning a Phase I/II multicenter clinical trial. Use of the drug is described generally, for example, in J.C. Kennedy et al., "Photodynamic Therapy (PDT) and Photodiagnosis (PD) Using Endogenous Photosensitization Induced by 5-Aminolevulinic Acid (ALA): Mechanisms and Clinical Results," Journal of Clinical  
15 Laser Medicine & Surgery, Vol. 14, No. 5, 1996, pp. 289-304, and E.W. Jeffes, "Photodynamic Therapy of Actinic Keratosis With Topical 5-Aminolevulinic Acid," Arch Dermatol, Vol 133, June 1997, pp. 727-732, both references herein entirely incorporated by reference.

When applied to tissue in low concentrations, cells treated with a PDT drug  
20 fluoresce. This allows for the detection (photodiagnosis) of atypical or cancerous cells. When applied in high concentrations, the drug can kill atypical cells. The articles by Kennedy et al. listed above suggest that a topical solution with a 20% concentration of Levulan, applied to non-melanoma skin and head and neck cancers, may be used for photodiagnosis. Jeffes et al. suggests that topically based solutions with PDT  
25 concentrations of 10 to 30% may be used to treat face and scalp lesions.

Applicants note that a PDT, such as Levulan, may also be utilized successfully with the present system 10 shown in FIG. 1 or the system 204 shown in FIG. 6. More particularly, Levulan may be used for detection and treatment of atypical or cancerous cells in the gynecological tract. In low concentrations, the drug may be used for applications  
5 such as for the detection, visualization, localization, screening, and diagnosis of atypical and cancerous cells.

Aside from such diagnostic applications, Applicants believe that PDT drugs may also be used for therapeutic applications in the gynecological tract. For example, by applying the drug in generally high concentrations, the drug can treat lesions in the  
10 gynecological tract. Therefore, in one embodiment, the PDT drug is used both to assist with diagnosis of atypical or cancerous tissue in the gynecological tract, as well as the treatment of that tissue.

As previously discussed, the photodynamic therapy agents can be applied via a number of different methods. FIG. 5 illustrates an alternative embodiment for applying a  
15 photodynamic probe and shows an endoscope examining tissue 5. The endoscope 505 shown in FIG. 5 may be used for applying photodynamic agents, as well as assisting with an examination of tissue fluorescence. As shown in FIG. 5, the endoscope includes a sheathing member 505, an application mechanism 525, and a catheter 530. Typically, the photodynamic agent is applied via the endoscope 505 having an application mechanism  
20 525. The application mechanism may be, for example, a brush, sponge, or similar application device. Brushes for such purposes ("cytobrushes") are readily available from a variety of distributors, such as, for example, Andwin Scientific of Canoga Park, California; Globe Scientific of Paramus, New Jersey; IMEB Inc. of San Marcos, California; Medical Packaging Corp. of Camarillo, California; Medscand (U.S.A.) Inc. of Hollywood, Florida;  
25 Shandon Lipshaw, of Pittsburgh, Pennsylvania; and Surgipath Medical Industries of

Richmond, Illinois. The endoscope 305 may be used to assist in locating a suspect area of tissue 5 and then used to apply an agent once the suspect tissue located or is identified.

Regardless of the mechanism used for diagnosis and treatment of atypical or cancerous tissue, the specific areas of the vaginal canal screened and treated may be  
5 monitored and recorded by a data collection device. Accordingly, the operator may have, for example, documentation that the entire relevant area has been previously reviewed, along with a record of any fluorescence detection. A record of how such tissue areas were treated can also be saved..

By monitoring and recording the area where fluorescence was noted during a  
10 previous examination, a physician may know more easily later where supplemental treatment should occur. Such supplemental treatment could include the application of a highly concentrated solution of a PDT drug. Similarly, by monitoring and recording the location of an agent application, for example, a physician may document treatment of the area. A physician may therefore, during a supplemental investigation, return an  
15 investigating device, such as the endoscopes shown in FIG. 1 or FIG. 5, to the same suspect tissue area for follow-up treatment.

In an alternative embodiment, the endoscope 535 is provided with a single diagnostic and therapeutic device. In this embodiment, the drug used with the present system 10 is applied via the single device. The single device is used both to collect tissue  
20 samples and is used to apply the agent. The endoscope 535 may be either is flexible or rigid. Whether flexible or rigid, however, a preferred embodiment utilizes a steerable endoscope. The device is remotely controlled from one end of the endoscope (or the catheter 530) after inserting the other end into the endo-cervical canal.

The application mechanism 525 may contain a brush that can be used for collection  
25 of suspect tissue, as well as, for example, a brush or sponge to topically apply the agent.

Again, the agent may be applied in varying concentrations. In low concentrations, the agent stimulates fluorescence of atypical or cancerous sell. In a high concentration, the agent kills atypical or cancerous cells. The endoscope may comprise a number of different types of tips to facilitate a variety of different applications for the same endoscope.

5 In still another alternative embodiment of the present invention, the endoscope comprises a plurality of endoscopic tubes. One of the tubes may contain the fiber optics coupled to a light source. Other tubes could contain a catheter. The tissue sample could then be removed from the patient by way of one of the endoscopic tubes. In this manner, the sample or multiple tissue samples could be taken from the same patient.

10 In the system shown in FIG. 1, the endoscope 10 is preferably coupled to the computing device 110. The computing device 100 monitors and records both (1) the diagnostic procedures taken by a physician when examining the gynecological tract for luminescent tissue, and (2) the therapeutic procedures taken by a physician in apply a higher concentration of the agent to tissue deemed atypical or cancerous. The endoscope supplies  
15 data to the device 110 for recording these procedures taken by the physician. In this manner, an archive is established regarding the tissue areas that the physician analyzed for luminances and which areas were detected as being atypical as atypical or cancerous. Such data may then be recalled during supplemental investigations, thereby directing the physician to the same areas, so that a treatment drug may be applied to the appropriate  
20 tissue. The system shown in FIG. 6 operates in a similar manner.

In an alternative embodiment, the system 10 includes a direction-indicator. For example, the computing device 110 shown in FIG. 1 includes a direction-indicator 212. The direction indicator directs the person operating the endoscope how to manipulate the sterile endoscope 30 so as to return the distal end or tip 32 of the endoscope 30 to a  
25 particular tissue area.. This area may be an area that was previously noted by the physician

as luminescing, thereby identifying atypical or cancerous cells. The system shown in FIG. 6 can operate in a similar manner.

Alternatively, the tissue area may have been previously noted as an area that had been treated (with, for example, a high concentration solution of the PDT drug) and the  
5 direction indicator 212 assists the physician in returning to the same area for further treatment to confirm that the treatment was successful.

Therefore, a data collection device, such as the computing device, records the location of the tissue that fluoresces during an initial examination. Furthermore, recordation provides the investigating physician documentation as to the tract location. Recordation  
10 also verifies that the physician investigated all suspect areas identified via the agent.

In an alternative embodiment, the endoscope 30 contains a position locator or position mechanism 55. (FIG. 1) The position mechanism provides for determining the absolute and/or relative XYZ location coordinates of the endoscope 30. This information can then be used by the data collection device for medical recording documentation  
15 localization during screening and diagnostics and relocation of endoscope and drug delivery systems for patient therapy.

The position mechanism 55 may be, for example, a position locator with a Radio Frequency emitter. The emitter may be located at the distal end 32 of the endoscope. The distal end or endoscope tip 32 may be replaceable.

20 The electrical power for the system may be bundled with the fiber optics of the endoscope. Preferably, the position mechanism is a XYZ spatial coordinate sensor. The position mechanism allows the endoscope 30 and related sensor to determine the absolute and/or relative location of the endoscope distal end within the canal. By noting the position of the endoscope tip while the tip is adjacent particular areas within the canal (such as areas  
25 that luminesce or areas which are undergoing treatment), the coordinates may be recalled by



the physician at a later time. This allows the physician to return the endoscope tip to the same area at a later for further analysis and treatment.

The position mechanism 55 provides a number of advantages. For example, it provides a reference point to return to within the gynecological tract for further investigation and reapplication of a drug. This allows the investigating physician to return to applied area to determine if the suspect cells are still atypical.

In still another alternative embodiment, the endoscope contains a sampling device. As previously discussed with respect to FIG. 2, the sampling device can contain a plurality of tissue samples taken from suspect or non-suspect areas. These samples may, for example, be extracted through a catheter of an endoscope such as the catheter 530 shown in FIG. 5.

Preferably, the sampling device places the tissue in a sample container having a plurality of compartments, divisions, sections, or the like. The compartments facilitate the isolation of various tissue samples. Therefore, a number of different samples from the same or different patients may be taken without getting the samples mixed with one another.

The container also includes a log for identifying from whom the sample was taken. The log could also identify the exact location from where within the patient the sample was taken. These various samples may then be individually analyzed.

Thus, the present system allows a physician to (1) image the tissue, (2) induce fluorescence, (3) provide image data to an analytical instrument, including a computer system, and (4) collect site-specific tissue samples in a way that is accurate, repeatable, and efficient. The system for diagnosis and treatment does not significantly increase the degree of invasiveness. The system also reduces the time for examining and taking of tissue samples, as compared to conventional Pap tests.

Since the tip 32 of the endoscope may be sterilized or may be replaced, the tip 32 can be adapted for use even in a practitioner's office. The system supports examination of the entire cervix and vagina, and is usable for the large majority of patients undergoing Pap tests. The system is relatively low cost and is easy to use by a single operator. The operator  
5 may confirm visually that all atypical or cancerous areas have been noted or sampled. The equipment used may be mass produced and is compatible with most of the relevant agents.

Further, the monitoring and recordation device allows users to image, detect, localize, quantitatively characterize, and visualize atypical or cancerous tissues from the human gynecological tract. Further, the system documents and records the areas reviewed  
10 by the user when diagnosing the patient. The system also records and documents the actions taken in treating the patient with, for example, a PDT drug.

While the invention has been described in conjunction with the presently preferred embodiments of the invention, persons of skill in the art will appreciate that variations may be made without departure from the scope and spirit of the invention. This true scope and  
15 spirit is defined by the appended claims, as interpreted in light of the foregoing.

**WE CLAIM:**

1. An apparatus for imaging tissue using autofluorescence, comprising:
  - an endoscope having a distal and proximal end;
  - a light source;
  - 5 a light processing unit located at a proximal end of the endoscope, wherein the light processing unit accepts light in a spectral region from the light source and directs the light through the endoscope to the tissue located at the distal end of the endoscope, wherein the light excites autofluorescence in the tissue;
  - optics located in the distal end of the endoscope which collect the excited
  - 10 autofluorescence in a spatially resolved manner and routes it to the light processing unit which separates the emitted light from the reflected light; and
  - a plurality of imaging detectors which receive the emitted light routed by the light processing unit, each imaging detector responsive to light in a different spectral band,
  - wherein an image is generated according to a plurality of independent
  - 15 measurements.
2. An apparatus for imaging tissue using autofluorescence, comprising:
  - an endoscope having a distal and proximal end;
  - a light source;
  - 5 a light processing unit located at a proximal end of the endoscope, wherein the light processing unit accepts light in a spectral region from the light source and directs the light through the endoscope to the tissue located at the distal end of the endoscope, wherein the light excites autofluorescence in the tissue;

optics located in the distal end of the endoscope which collect the excited  
10 autofluorescence in a spatially resolved manner and routes it to the light processing unit  
which separates the emitted light from the reflected light; and

an imaging detector which receives the emitted light routed by the light processing  
unit, the imaging detector being responsive to light in a different spectral band,

wherein an image is generated according to a plurality of independent  
15 measurements.

3. An apparatus for sampling tissue, comprising:

an endoscope device having a sampling device, the sampling device capable of  
taking a plurality of tissue samples; and

5 a plurality of captive units in communication with the sampling device;  
such that each tissue sample is deposited in a separate captive unit.

4. A method for diagnosing atypical tissue in the human gynecological tract  
comprising, in combination:

applying a photodynamic agent to said tract;

5 allowing said photodynamic agent to be absorbed by said tissue;

inserting a diagnostic tool into said tract, said tool including a light source and a  
monitor; and

observing florescence of tissue within said tract.

5. A diagnostic tool for use by an operator in diagnosing atypical and cancerous tissue in the human gynecological tract comprising, in combination:
- a endoscope having a tip, said tip defining a position;
  - 5 said tip including a light source, position locator and a fluorescence detector, said position locator providing position signals regarding said position of said tip, and said fluorescence detector providing fluorescence signals regard fluorescence of tissue near said tip; and
  - a data collection device for receiving said position signals and fluorescence signal and
  - 10 providing information to said operator regarding said position of said tip and fluorescence of said tissue in said tract.

6. A diagnostic tool as claimed in claim 5 wherein said endoscope is steerable by said operator.

7. A therapeutic tool for use by an operator in treating atypical and cancerous tissue in the human gynecological tract comprising, in combination:
- an endoscope having a tip, said tip defining a position;
  - 5 said tip including a light source, position locator and a fluorescence detector, said position locator providing position signals regarding said position of said tip, and said fluorescence detector providing fluorescence signals regard fluorescence of tissue near said tip; and

a data collection device for receiving said position signals and florescence signal and  
10 recording said position signals as a recorded position when said tip is near fluorescing  
tissue;

said data collection device further providing information to said operator regarding  
said position of said tip relative to said recorded position, whereby said operation may  
return said tip to said recorded position for further treatment.

8. The endoscope of claim 7 wherein the endoscope is steerable.

9. The of claim 7 wherein the endoscope further comprises an application  
mechanism for applying a probe onto tissue.

10. The tool of claim 7 wherein the endoscope further includes a sampling  
device to collect tissue samples and to apply a high concentration of a photodynamic drug to  
the tissue.

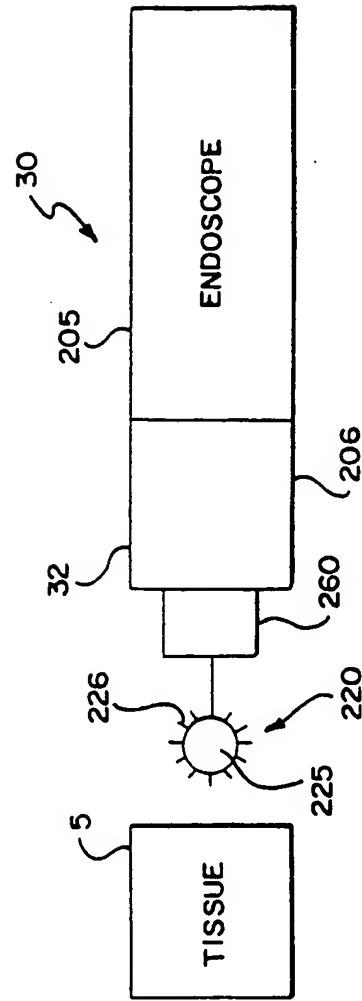
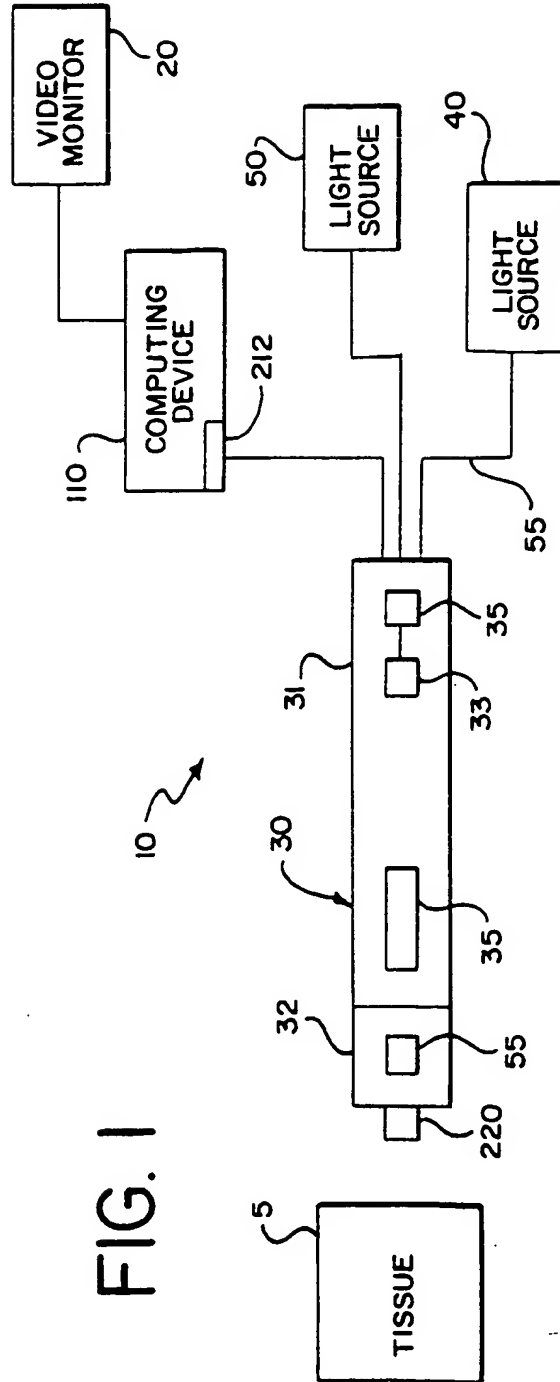
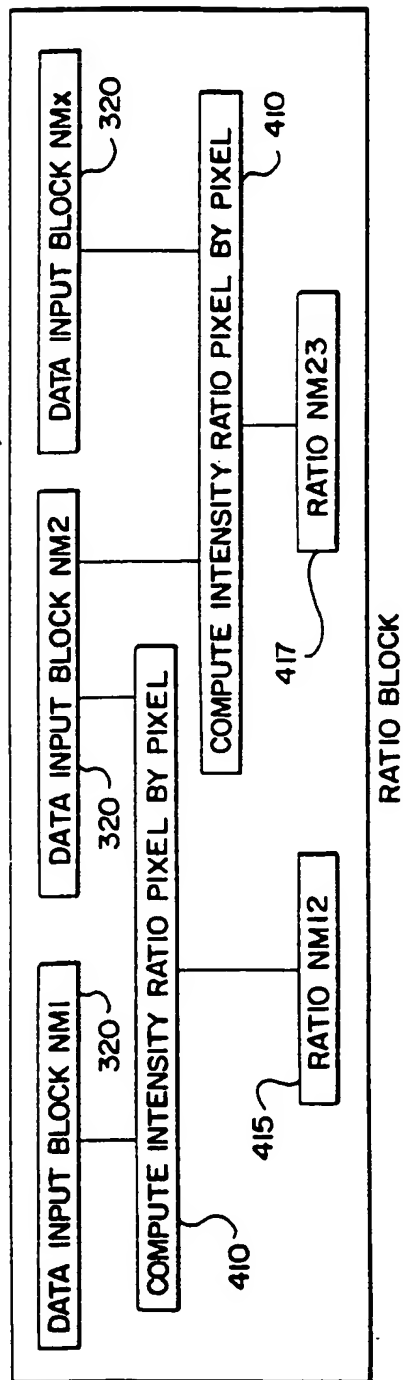
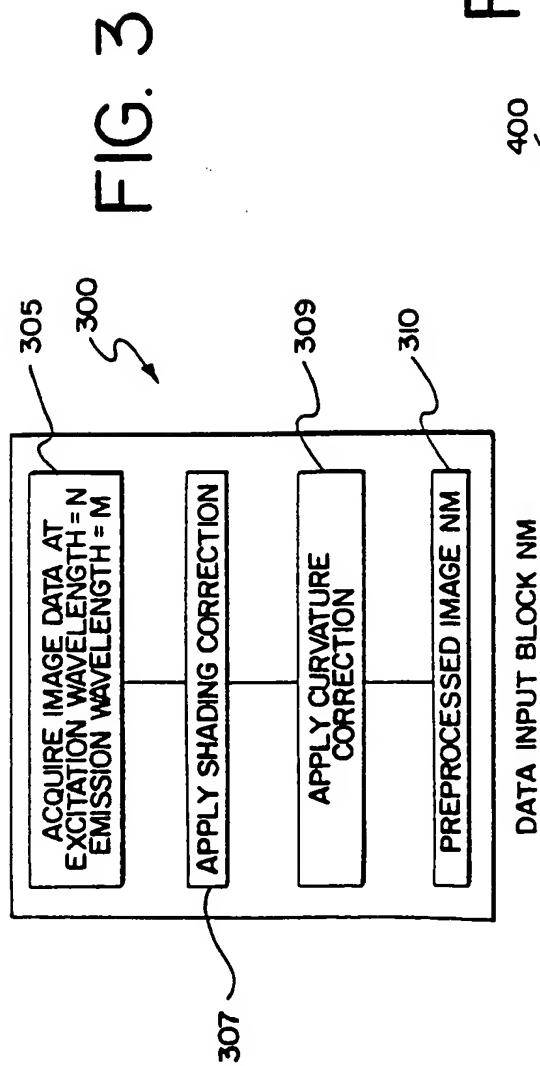


FIG. 1

FIG. 2

2/3





3/3

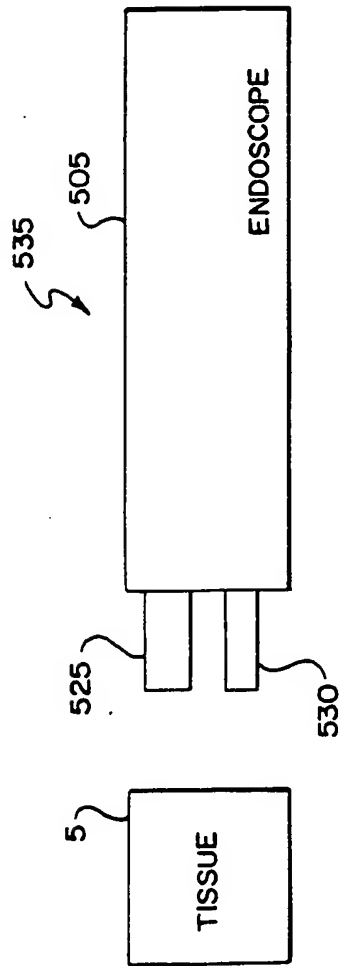


FIG. 5

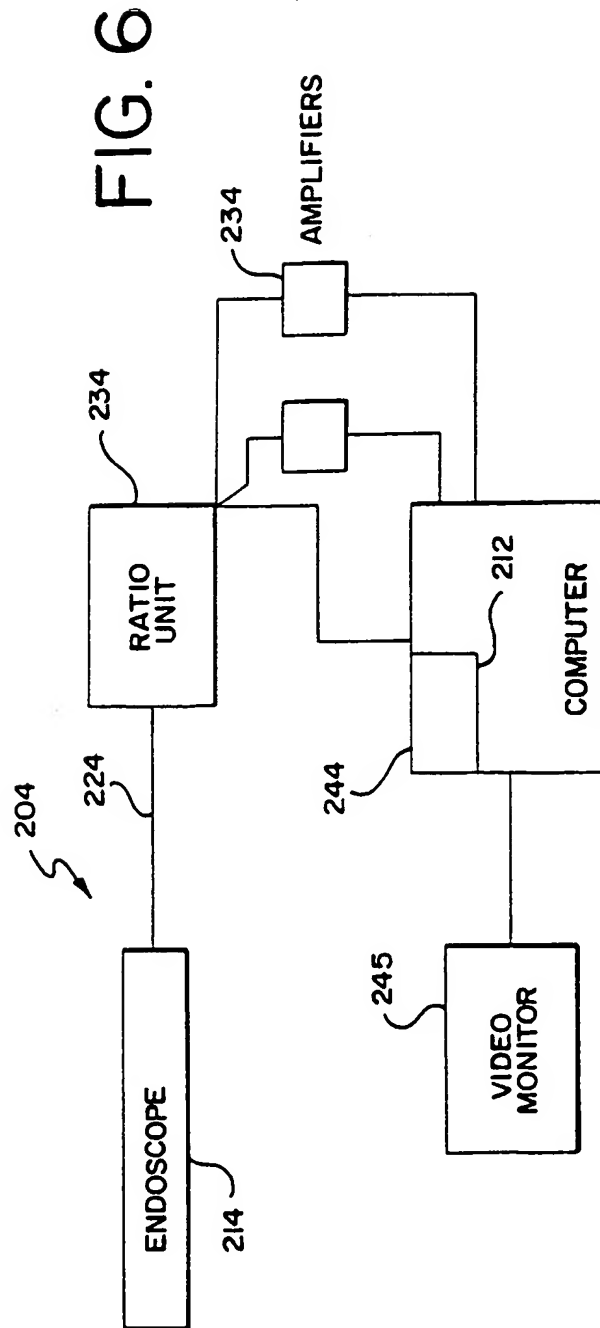


FIG. 6

# INTERNATIONAL SEARCH REPORT

International Application No  
PCT/US 98/21710

**A. CLASSIFICATION OF SUBJECT MATTER**  
IPC 6 A61B5/00

According to International Patent Classification (IPC) or to both national classification and IPC

**B. FIELDS SEARCHED**

Minimum documentation searched (classification system followed by classification symbols)  
IPC 6 A61B

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

**C. DOCUMENTS CONSIDERED TO BE RELEVANT**

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X X Y	US 4 905 670 A (E.L. ADAIR ) 6 March 1990 see column 2, line 3 - line 49 see column 3, line 39 - column 4, line 64 ---	1,2,4,5 7 6,8
X Y A	US 4 718 417 A (C. KITTRELL ET AL.) 12 January 1988 see column 24, line 4 - line 55 see column 25, line 62 - column 26, line 28; figures 4A,24 ---	1,2 6,8 5,7,9
X	WO 96 28084 A (UNIVERSITY OF TEXAS) 19 September 1996 see page 2, line 30 - page 3, line 32 see page 9, line 12 - page 12, line 17 --- -/--	1-3,5

☒ Further documents are listed in the continuation of box C.

☒ Patent family members are listed in annex.

\* Special categories of cited documents :

- "A" document defining the general state of the art which is not considered to be of particular relevance
- "E" earlier document but published on or after the international filing date
- "L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)
- "O" document referring to an oral disclosure, use, exhibition or other means
- "P" document published prior to the international filing date but later than the priority date claimed

- "T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
- "X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
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# INTERNATIONAL SEARCH REPORT

International Application No  
PCT/US 98/21710

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT		
Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	US 5 623 932 A (N. RAMANUJAM ET AL.) 29 April 1997 see column 3, line 56 - column 4, line 22 see column 6, line 12 - line 57 see column 7, line 12 - column 8, line 15 ---	1,2,5
A	US 5 596 992 A (D.M. HAALAND ET AL.) 28 January 1997 see column 6, line 10 - column 7, line 13 -----	1-3,5,7, 9

# INTERNATIONAL SEARCH REPORT

Information on patent family members

Inter: nat Application No

PCT/US 98/21710

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
US 4905670 A	06-03-1990	CA 1322591 A DE 68924784 D DE 68924784 T EP 0451200 A WO 9007299 A US 5026368 A US 5143054 A	28-09-1993 14-12-1995 30-05-1996 16-10-1991 12-07-1990 25-06-1991 01-09-1992
US 4718417 A	12-01-1988	CA 1279901 A CA 1339056 A CA 1317641 A CA 1329655 A DK 130586 A FI 861209 A JP 2589674 B JP 61257638 A JP 2739933 B JP 9117407 A	05-02-1991 29-07-1997 11-05-1993 17-05-1994 23-09-1986 23-09-1986 12-03-1997 15-11-1986 15-04-1998 06-05-1997
WO 9628084 A	19-09-1996	US 5697373 A CA 2190374 A EP 0765134 A JP 10505167 T	16-12-1997 19-09-1996 02-04-1997 19-05-1998
US 5623932 A	29-04-1997	US 5421339 A AU 6946894 A CA 2162922 A EP 0702526 A JP 8511179 T WO 9426168 A	06-06-1995 12-12-1994 24-11-1994 27-03-1996 26-11-1996 24-11-1994
US 5596992 A	28-01-1997	NONE	